

Genetic analysis of agronomic and fibre traits using four interspecific chromosome substitution lines in cotton

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Abstract

Backcrossed chromosome substitution lines (CS-B) have been developed with a homologous pair of chromosomes or chromosome arms of *Gossypium barbadense* (3-79) germplasm substituted for the homologous *Gossypium hirsutum* (TM-1) chromosomes or chromosome segments. We report on agronomic and fibre trait performance of four backcrossed chromosome or chromosome arm substitution lines including chromosomes 01, 11sh (chromosome 11 short arm), 12 sh and 26 Lo (chromosome 26 long arm). Data for agronomic and fibre traits were collected from replicated field experiments at two different locations in 2 years, and analysed under an additive dominance genetic model. CS-B 12sh had higher, while CS-B 01 and CS-B 26Lo had lower boll weight than TM-1. The presence of significant negative additive effects for micronaire with CS-B 01 and significant positive additive effects for elongation and fibre strength with CS-B 11sh suggested the substituted chromosome arms of 3-79 in these CS-B lines were more likely carrying genes causing these effects. Results revealed that several CS-B lines had significant homozygous and heterozygous dominance effects for different agronomic and fibre traits suggesting that specific CS-B lines may be useful for improving agronomic and fibre traits in hybrid cottons. These CS-B lines also provide novel genetic resources for improving upland cotton germplasm.

Key words: chromosome substitution lines — cotton — fibre traits — agronomic traits

Cotton is the world's most leading natural textile fibre and an important oilseed crop. It is cultivated on about five million hectares per year, more than any crop except maize, wheat, or soybean (USDA-NASS 2002). It is one of the premier industrial crops in the U.S., capturing 57% of the domestic textile market (Cotton Incorporated 2006). Domestic yarn and textile manufacturers have adopted high speed spinning and yarn technologies. These new yarn and textile production technologies place greater strain on the cotton fibre than the older textile manufacturing technology and thus, there is a demand for cottons with improved fibre qualities, especially strength and uniformity.

Genetic resources are the key to ensuring that future demands of superior fibre qualities, production and profits in the world cotton market can be met. The exceptional fibre length, strength and fineness of Pima and Sea Island cotton (*Gossypium barbadense* L.) provide superior spinning and manufacturing performance and confer a price advantage over the more widely grown upland cotton (*Gossypium hirsutum* L.). However, upland cotton occupies over 90% of the cotton-growing area in the world because of its higher yield and wider

adaptation. Yield and fibre properties of USA upland cottons have been on a plateau for the last decade (Cantrell et al. 2003, Cotton Incorporated 2006). A 'Blue Ribbon Committee' organized by the National Cotton Council reported that cotton yield in USA had peaked in 1992, but decreased at a rapid rate of about 16.8 kg/ha/year or 3.3% annually (Helms 2000).

Despite the economic importance of upland cotton, the genetic base of upland cultivars and elite germplasm is very narrow (Bowman et al. 2003). Domestication of cotton most likely originated from a small subset of the wild genotypes (Gingle et al. 2006). However, the germplasm base of *Gossypium* is very diverse with over 50 species including six tetraploid species. One of the solutions to the problem of genetic vulnerability is interspecific introgression to increase the genetic diversity. Interspecific introgression from *G. barbadense* is an attractive strategy to improve fibre qualities in upland cotton, especially considering the superior fibre properties of *G. barbadense*. However, *G. barbadense* has failed to be used widely as a source of diversity by upland breeders because of the relative difficulties and inadequacy results from the conventional methods of interspecific introgression (Stephen 1949, McKenzie 1970). Problems included poor agronomic qualities of the progeny, distorted segregation, sterility, mote formation and limited recombination because of incompatibility between the genomes. A majority of interspecific crosses between *G. hirsutum* and *G. barbadense*, using conventional methods, did not produce economically useful fibres (Rong et al. 2004, Lacape et al. 2005). It has been well documented that widespread deficiencies occurred in donor chromatin (*G. barbadense*) in the interspecific backcross population (Jiang et al. 2000). An alternative approach would be to use alien chromosome substitution lines.

Recently, we have developed a series of 17 backcrossed chromosome substitution lines (CS-B), is each of which a homologous pair of chromosomes or chromosome arms from *G. barbadense* (Pima 3-79) germplasm has been substituted for the homologous *G. hirsutum* (TM-1) chromosomes or chromosome arms (Stelly et al. 2005). These lines provide an ideal opportunity to associate important fibre and agronomic traits with an individual chromosome or chromosome arm (Saha et al. 2004, 2006). These CS-B euploid lines are also useful as parents to introgress favourable genes from specific *G. barbadense* chromosomes and to develop improved upland cultivars (Jenkins et al. 2006, 2007).

Previously, we reported an investigation of fibre and agronomic traits of 13 released CS-B lines either in their parental or hybrid background (Saha et al. 2004, 2006). Here we report on agronomic and fibre trait performance of four additional euploid ($2n = 52$) backcrossed chromosome substitution lines. The association of important agronomic and fibre traits with four chromosomes of *G. barbadense* based on comparative analysis of these CS-B lines using the chromosome-specific F_2 hybrids, parental CS-B lines, the recurrent TM-1 and donor 3-79 parent lines, respectively, are reported. Agronomic and fibre trait performance levels of these CS-B lines across four environments in replicated plots in each environment are provided. This study was designed to evaluate the effect of a chromosome or chromosome segment from *G. barbadense* on fibre and agronomic traits in the *G. hirsutum* genome and will provide useful genetic information for future cotton breeding and genetic studies.

Materials and Methods

Four near-isogenic BC_5S_1 chromosome substitution lines containing pairs of different *G. barbadense* chromosome or chromosomes arm or segments (CS-B lines), were crossed with TM-1 (*G. hirsutum*) and 3-79 (*G. barbadense*) for agronomic and fibre trait evaluation. The inbred TM-1 was developed from the commercial variety 'Deltapine 14' and inbred over 40 generations by self pollination, and is considered as the

primary genetic standard for cotton genetics, cytogenetics and for molecular standard for *G. hirsutum* (Kohel et al. 1970, 2001). Line 3-79 originated as a double haploid from *G. barbadense* (Endrizzi et al. 1985). In each CS-B line, a single pair of chromosome or chromosome arms of TM-1 had been replaced by the corresponding chromosomal segment of the 3-79 genome (Fig. 1). BC_5S_1 seeds from euploid ($2n = 52$) CS-B lines specific to four different chromosomes or chromosome segment were used in crossing with TM-1 to develop chromosome-specific F_2 hybrids. Development of these CS-B lines has previously been reported (Saha et al. 2004, Stelly et al. 2005).

We used the parental CS-B 26Lo line in our previous study (Saha et al. 2006); however, because of lack of sufficient F_2 seed supply the chromosomal effect was estimated without including the F_2 hybrid lines. Chromosome effect of 26Lo was analysed and included both parental and chromosome-specific F_2 hybrid lines in the current results. In addition, we report the effect of alien chromosomes 01, 11sh, 12sh, respectively, on agronomic and fibre traits in this report.

The F_2 hybrids and parental lines were planted in a randomized complete block design with four replications at two locations (four environments) in 2004 and 2005 at Mississippi (MS). Soil types of the two locations were Marietta loam (fine-loamy, siliceous, active, fluvaquent Eutrudepts) type and a Leeper silty clay loam (fine, smectitic, non-acid, thermic Vertic Epiaquept) type, respectively. Standard agronomic practices were followed during the growing season of all environments.

In 2004, plots consisting of a single row 0.97 m wide by 12 m long were planted on 13 May and harvested on 29 October at both locations. In 2005, plot size at one location was identical to that in

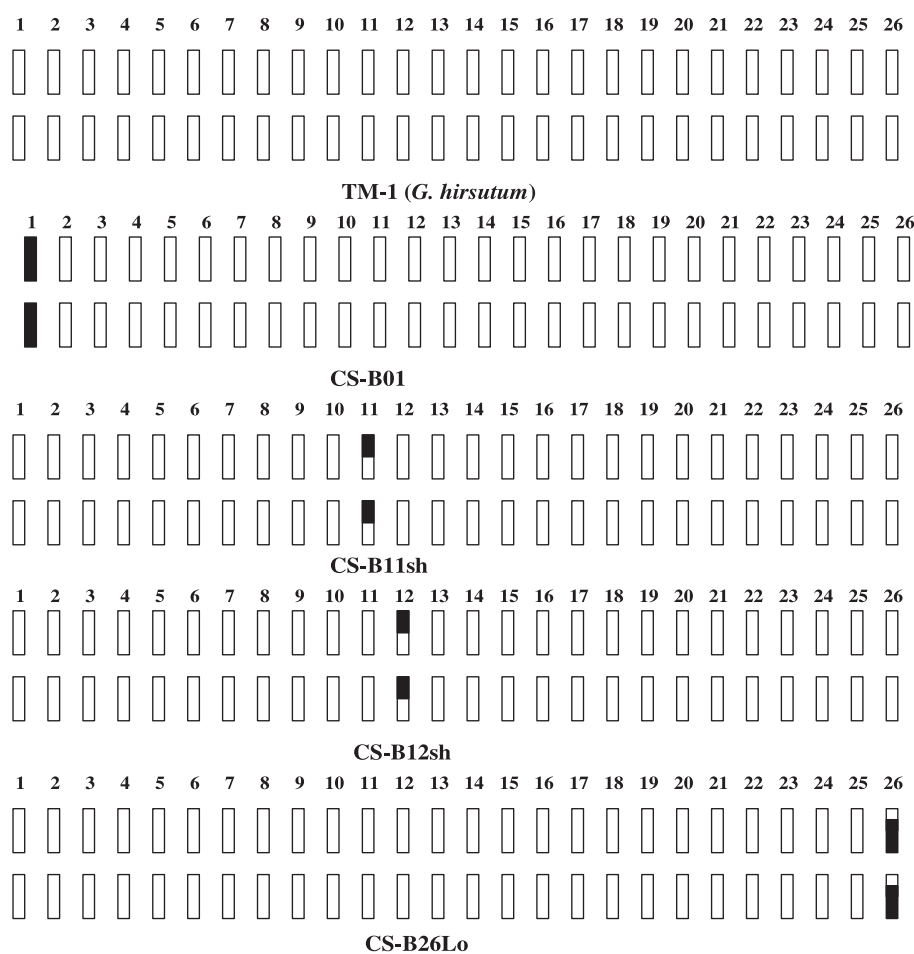


Fig. 1: Diagram of the genomes of TM-1 (*Gossypium hirsutum*) and four different euploid chromosome substitution lines showing the introgression of an alien chromosome or chromosome arm (black colour) from 3-79 (*Gossypium barbadense*) in the respective backcrossed chromosome substitution lines (CS-B) line

2004. Plots were planted and harvested on 12 May and 14 October, respectively. At the second location, the plots consisted of a single row 0.97 m wide by 9 m long and were planted on 13 May and harvested on 24 October. All experiments were carried out at the Plant Science Research Center, Mississippi State, MS.

A 25-boll sample was hand harvested from first position bolls near the middle nodes of plants in each plot prior to machine harvest. Boll samples were weighed and ginned on a 10-saw laboratory gin, after which the lint and seed fractions were weighed and lint percentage (LP) calculated by dividing lint fraction by total weight of seed and lint. Fibre samples were evaluated by STARLAB, Inc., Knoxville, TN, for micronaire (MIC), 2.5% span length (SL2.5), elongation (EL) and fibre strength (T1) using high volume instrument equipment. Lint yield (LY) was estimated by multiplying seed cotton yield (YLD) with LP.

Genetic models and statistical methods: All agronomic and fibre traits were subjected to analysis of variances and multiple comparisons using SAS (SAS Institute 2001). Means were separated using Fisher's protected least significant difference at the 0.05 level. An additive-dominance (AD) genetic model with $G \times E$ interaction was employed for genetic analysis (Zhu 1994, Tang et al. 1996, Jenkins et al. 2006, 2007, Saha et al. 2006). Variance components and genetic effects were calculated by the mixed linear model approach (Rao 1971, Zhu 1989, 1993).

A jackknife re-sampling technique was applied to calculate the standard error (SE) for each parameter by removal of one replicate within each environment (Wu et al. 2006). In this study, non-pseudo-value-based estimated values rather than pseudo-value-based estimates (Miller 1974) were used to calculate the SE for each parameter. Both methods provided similar estimates, but the non-pseudo-value-based jackknife provided greater precisions in this study. An appropriate *t*-test was used to detect significance of each parameter. A confidence interval test (95%) was utilized to estimate the significance of genetic effects between genotypes. Results on the chromosomal effects from both overall mean data and the deviation from TM-1 are presented. All genetic analyses were conducted using a program written in C++ by Wu et al. (2003).

Results

In our research, each CS-B line has a pair of chromosomes or arms of *G. hirsutum* inbred TM-1 replaced by the respective pair from *G. barbadense* doubled-haploid 3-79 (Fig. 1). These backcrossed chromosome substitution lines are near-isogenic to the recurrent parent TM-1 for 25 chromosome pairs and also similar to each other for 24 chromosome pairs (Stelly

et al. 2005, Saha et al. 2006). Such uniform genetic background provided an opportunity to compare these lines and detect the genetic effect of a chromosome or chromosome arm for a specific trait.

Mean comparisons for CS-B lines, chromosome-specific F₂ hybrids with donor and recurrent parental lines

Seed cotton yield, LY, LP, fibre MIC, length, T1 and EL of donor parent 3-79 were significantly different from the recurrent parent TM-1 (Table 1). Line 3-79, had poor agronomic traits compared with TM-1. However, as expected, 3-79 had the lower MIC and longer and stronger fibres compared with TM-1 (Table 1). The range of variation between TM-1 and 3-79 in agronomic and fibre traits supported the rationale for selecting these as parents for developing CS-B lines. CS-B 01, CS-B 11sh and CS-B 26Lo lines had significantly lower YLD and LY than TM-1. F₂ hybrids for these CS-B lines produced increased YLD and LY compared with their respective parental CS-B line. However, none of these four CS-B lines or their F₂ hybrid produced a significantly greater yield than TM-1. CS-B 12sh had heavier bolls, while CS-B 01 and CS-B 26Lo had lighter bolls than TM-1. The F₂ hybrids of CS-B 11sh \times TM-1 and CS-B 12sh \times TM-1 had greater BW than TM-1. Both CS-B 01 and CS-B 11sh had lower LP values than either TM-1 or 3-79. CS-B 01 had lower MIC, whereas, CS-B 11sh and CS-B 26Lo had higher MIC than TM-1. CS-B 01 and CS-B 26Lo had lower T1 than either 3-79 or TM-1. CS-B 11sh had a higher EL percentage than TM-1 and 3-79. The F₂ hybrid of CS-B 11sh \times TM-1 had higher EL percentage than TM-1. Fibre 2.5% span length was longer for CS-B 11sh and shorter for CS-B 12sh compared with TM-1. F₂ hybrids of CS-B 26Lo \times TM-1 had greater 2.5% fibre span length than TM-1.

Variance components

Variance components of all the traits were calculated and expressed as a proportion of the total phenotypic variance (Table 2). The proportion of residual variance ranged from 35.5% for EL percentage to 53.6% for LP. The relative importance of additive vs. dominance effects varied widely among the traits. All agronomic and fibre traits except YLD

Table 1: Mean values of fibre and agronomic traits of CS-B lines over four environments

Chromosome	Fibre and agronomic traits							
	SL (mm)	T1 (kNM/kg)	EL (%)	MIC	LP (%)	BW (g)	YLD (kg/ha)	LY (kg/ha)
01	29.53	271*	8.44*	4.34*	33.17*	4.88*	1821*	604*
11sh	29.99*	291	8.71*	4.76*	32.40*	5.27	1880*	609*
12sh	28.97*	292	8.21	4.69	34.51	5.51*	2168	746
26Lo	29.85	278*	8.12	5.08*	33.74	4.74*	1540*	519*
TM-1	29.59	290	8.21	4.58	34.04	5.26	2159	734
3-79	34.70	369	8.59	3.93	35.78	3.67	935	333
01 \times TM-1F2	29.53	284	8.23	4.39*	33.82	5.16	2095	707
11sh \times TM-1F2	29.83	291	8.50*	4.64	33.57	5.49*	1903*	638*
12sh \times TM-1F2	29.48	288	8.20	4.62	33.81	5.62*	2125	717
26Lo \times TM-1F2	30.08*	286	8.10	4.88*	33.66	5.31	2251	756
LSD0.05	0.37	7.4	0.17	0.14	0.49	0.20	241	84

CS-B, backcrossed chromosome substitution lines; SL, 2.5% span length; T1, fibre strength; EL, fibre elongation; MIC, micronaire; LP, lint percentage; BW, boll weight; YLD, seed cotton yield; LY, lint yield.

The bold font rows represent parental lines used in developing the chromosome substitution lines.

*Significantly different from TM-1 at the 0.05 probability level.

Table 2: Variance components expressed as proportions of the phenotypic variances for fibre and agronomic traits using parents, CS-B lines and F₂ populations

Variance component	SL (mm)	T1 (kNM/kg)	EL (%)	MIC	LP (%)	BW (g)	YLD (kg/ha)	LY (kg/ha)
V_A/V_P	0.152**	0.264**	0.233**	0.433**	0.065**	0.133 **	0.000	0.000
V_D/V_P	0.044**	0.015	0.000	0.004	0.325**	0.333 **	0.379**	0.369 **
V_{AE}/V_P	0.000	0.002	0.000	0.140 **	0.074**	0.065 **	0.128**	0.139 **
V_{DE}/V_P	0.345**	0.200**	0.412**	0.000	0.000	0.055 **	0.033	0.020
V_e/V_P	0.460**	0.519**	0.355**	0.423 **	0.536**	0.413 **	0.460**	0.472 **

CS-B, back crossed chromosome substitution lines; SL, 2.5% span length; T1, fibre strength; EL, fibre elongation; MIC, micronaire; LP, lint percentage; BW, boll weight; YLD, seed cotton yield; LY, lint yield; V_A , additive variance; V_D , dominance variance; V_{AE} , additive by environment variance; V_{DE} , dominance by environment variance; V_e , error variance; V_P , phenotypic variance.

**Significantly different than zero at the 0.05 and 0.01 probability level respectively.

and LY expressed significant additive effects ranging from 6.5% LP to 43.3% MIC. However, dominance effects exceeded additive effects for all of the agronomic traits ranging from 32.5% to 36.9%. Dominance effects suggest that these CS-B lines may have potential for hybrid cotton production which is a common practice in countries such as India and China. Both additive and dominance \times environment effects were significant for several traits.

Chromosome effects

The predicted values of genetic effects obtained from the AD genetic model were expressed as deviations from the respective population mean, not from the mean of TM-1. However, the information for significant deviation of any CS-B line from TM-1 is also provided (Tables 3 and 4).

The additive effects predicted for different CS-B lines varied for different traits (Table 3). Significant negative additive effects in SL were associated with CS-B 12sh and CS-B 01 lines, suggesting that the chromosome arms of 3-79 in TM-1 background were negatively associated with SL. However, chromosome 11sh and 26Lo had greater additive effects than TM-1 on SL, suggesting that the genes associated with these

chromosomes had positive additive effects on SL. We also observed that CS-B 01, CS-B 12sh and CS-B 26Lo had smaller additive effects than TM-1 for T1. These results indicate that 3-79 loci associated with these alien chromosomes significantly reduce T1, independently and/or through interactions. CS-B 01 and CS-B 11sh had higher additive effects while CS-B 26Lo had lower additive effect compared with TM-1 on EL, suggesting that genes affecting EL are likely associated with these two chromosome arms. The additive effects for MIC associated with CS-B 01 were negative and lower than TM-1 suggesting that this chromosome arm might carry genes reducing MIC. CS-B 01 and CS-B 11sh had lower additive effects for LP than TM-1, indicating that chromosome 1 and chromosome arm 11sh were associated with reduced LP. CS-B 01, CS-B 11sh and CS-B 26Lo had lower additive effects for BW than TM-1.

Homozygous and heterozygous dominance effects related to chromosome arms were determined by the comparative analysis of TM-1 with CS-B lines and their hybrids (Saha et al. 2006). Differences among the lines were attributed to the substituted chromosome pair based on the assumption that all other 25 pairs of chromosomes were in uniform TM-1 background in CS-B lines. The difference in homozygous

Table 3: Additive effects and standard errors for agronomic and fibre traits expressed as deviations from population grand mean

Chromosome	SL (mm)	T1 (kNM/kg)	EL (%)	MIC	LP (%)	BW (g)
01	-0.100 \pm 0.004*	-5.64 \pm 0.11*	0.031 \pm 0.002*	-0.184 \pm 0.002*	-0.056 \pm 0.005*	-0.130 \pm 0.003*
11sh	0.133 \pm 0.004*	3.04 \pm 0.07*	0.227 \pm 0.001*	0.030 \pm 0.001*	-0.235 \pm 0.016*	0.030 \pm 0.001*
12sh	-0.274 \pm 0.006*	2.32 \pm 0.09*	-0.053 \pm 0.003*	0.003 \pm 0.002*	0.168 \pm 0.012*	0.112 \pm 0.004
26Lo	0.176 \pm 0.006*	-2.86 \pm 0.09*	-0.122 \pm 0.001*	0.212 \pm 0.002*	0.008 \pm 0.003*	-0.125 \pm 0.004*
TM-1	0.065 \pm 0.003	3.14 \pm 0.07	-0.084 \pm 0.002	-0.061 \pm 0.002	0.116 \pm 0.009	0.113 \pm 0.003

SL, 2.5% span length; T1, fibre strength; EL, fibre elongation; MIC, micronaire; LP, lint percentage; BW, boll weight.

*Significantly different from TM-1 at the 0.05 probability level.

Table 4: Homozygous and heterozygous dominance effects and standard errors for agronomic and fibre traits expressed as deviations from population grand mean

	SL (mm)	T1 (kNM/kg)	MIC	LP (%)	BW (g)	YLD (kg/ha)	LY (kg ^{-ha})
Hom dom 01	0.011 \pm 0.004*	-0.109 \pm 0.041*	-0.001 \pm 0.001	-0.286 \pm 0.011*	-0.103 \pm 0.004*	-102.0 \pm 5.5*	-38.7 \pm 1.9
Hom dom 11sh	0.038 \pm 0.006*	0.020 \pm 0.011	0.002 \pm 0.001	-0.580 \pm 0.012*	-0.110 \pm 0.005*	88.6 \pm 5.5*	19.1 \pm 1.7*
Hom dom 12sh	-0.115 \pm 0.017	0.077 \pm 0.029*	0.001 \pm 0.001	0.569 \pm 0.016*	-0.057 \pm 0.005*	97.9 \pm 8.3*	44.9 \pm 2.9*
Hom dom 26Lo	-0.086 \pm 0.014	-0.051 \pm 0.019*	0.003 \pm 0.002	0.199 \pm 0.011	-0.305 \pm 0.007*	-404.1 \pm 7.1*	-131.3 \pm 2.4*
Hom dom TM1	-0.115 \pm 0.019	0.001 \pm 0.006	0.000 \pm 0.000	0.164 \pm 0.022	-0.376 \pm 0.010	-119.1 \pm 7.5	-34.7 \pm 2.5
Het dom 01	-0.074 \pm 0.013	0.104 \pm 0.040	-0.016 \pm 0.010	0.444 \pm 0.024*	-0.023 \pm 0.010*	142.5 \pm 13.3*	54.7 \pm 4.6*
Het dom 11sh	-0.005 \pm 0.006*	0.019 \pm 0.010	-0.002 \pm 0.001	0.628 \pm 0.023*	0.273 \pm 0.012*	-300.6 \pm 10.5*	-89.3 \pm 3.4*
Het dom 12sh	0.083 \pm 0.015*	-0.110 \pm 0.042*	-0.002 \pm 0.001	-0.758 \pm 0.031*	0.312 \pm 0.014*	-110.6 \pm 16.2	-53.5 \pm 5.7*
Het dom 26Lo	0.264 \pm 0.040*	0.048 \pm 0.020	0.013 \pm 0.007	-0.380 \pm 0.022*	0.389 \pm 0.013*	707.3 \pm 16.0*	228.8 \pm 5.4*

SL, 2.5% span length; T1, fibre strength; MIC, micronaire; LP, lint percentage; BW, boll weight; YLD, seed cotton yield; LY, lint yield.

*Significantly different from TM-1 at the 0.05 probability level.

dominance effects between a CS-B line and TM-1 is estimated from the difference in effects between the substituted chromosome pair and the respective pair of TM-1 chromosomes. The heterozygous dominance effects of a specific substituted chromosome are estimated from the difference in the effects of bulk F₁ and TM-1. Homozygous and heterozygous dominance effects are summarized in Table 4.

All CS-B lines and crosses had dominance effects that deviated significantly from the population means for all the agronomic traits (Table 4). Numerically, the homozygous vs. heterozygous dominance effects had opposite signs for the majority of the traits, suggesting that dominance differences were either because of interactions of alleles in the same chromosome pair or with alleles in other chromosome pairs. Results indicated that CS-B 01 had positive homozygous and negative heterozygous dominance effects on 2.5% SL. CS-B 01 also had negative homozygous dominance effect on T1, suggesting that genes associated with this alien chromosome would reduce T1 in the TM-1 genetic background.

Discussion

Published research has provided valuable information on additive effects, dominance effects (Cockerham 1980) and their G × E interaction effects (Zhu 1994) for a specific trait at the whole genome level using conventional methods of crosses between two diverse parents in cotton. The genetic background of the CS-B lines provided an opportunity to dissect the effect of an individual chromosome or arm under three different conditions: (i) the homozygous condition (euploids) in the TM-1 genetic background, (ii) the homozygous condition in the 3-79 genetic background and (iii) the F₂ generation which is segregating for the TM-1 and 3-79 alleles on the specific substituted chromosome or chromosome arm. For a specific CS-B line the difference in genetic effects between the line and TM-1 could be because of genes on the specific substituted chromosome and/or because of epistasis between TM-1 genes and genes on the substituted chromosome (Saha et al. 2006). The genetic background of some of the original aneuploid lines was not TM-1. However, these were backcrossed multiple times with TM-1 to recover the hypoaneuploid TM-1 near isogenic parents used to breed the CS-B lines. The possibility exists that some remnants of the genome in which the hypoaneuploid was originally discovered is present in the TM-1 near-isogenic stock, despite repeated backcrossing. This could have contributed to some of the trait-chromosome associations detected. Similarly, there is also a possibility that the observed genetic effects could have been due to some unlinked residual effect of *G. barbadense* chromatin in other chromosomes and is independent of the homozygous condition of the substituted chromosome or arm from *G. barbadense* (Saha et al. 2004, 2006). Any genetic change resulting from mutation could be another remote possibility that might have confounded the results. Lines similar to CS-B were used as a powerful tool in QTL analysis and germplasm improvement in other crops including wheat and tomato (Campbell et al. 2003, 2004, Canady et al. 2005).

CS-B F₂ analyses are relatively free of the interactions of inter-chromosomal loci of the chromosomes other than the substituted chromosome because of the complete replacement of 25 alien species chromosomes with the recurrent parent TM-1 chromosomes out of the 26 pairs of chromosomes (~96% of genome). This provides a unique opportunity to

detect novel allelic effect of a chromosome for a specific QTL by reducing the genetic noise from the genetic interactions of other chromosomes that normally arises in conventional interspecific F₂ populations. In a previous study, we showed that although the 3-79 parental line produced a low number of flowers, introgressing the short arm of chromosome 5 into TM-1 allowed us to detect a novel allele(s) which increased the flower number significantly above the parents TM-1 and 3-79 (McCarty et al. 2006). CS-B 26Lo expressed a significant positive heterozygous dominance effect on YLD and LY, suggesting that the novel alleles associated with the alien chromosome 26Lo might be useful in the yield improvement for countries developing hybrid cotton. Given the nature of multi-genic traits and their epistatic effect on yield, it seems reasonable that individual 3-79 alleles or QTLs will be discovered to improve yields.

Many of the agronomic and fibre traits in cotton are affected by quantitative traits controlled by genes with major and minor effects (Rong et al. 2004, Lacape et al. 2005, Ulloa et al. 2005, Zhang et al. 2005). CS-B lines will be useful for detecting these genetic effects associated with the alien chromosomes. The strategy utilized in this work and prior research (Jenkins et al. 2006, 2007, Saha et al. 2006, Wu et al. 2006) allowed us to partition the chromosome specific genetic effect into additive and dominance components for each trait. We detected some novel alleles associated with the alien chromosome one (CS-B 01), chromosome 12 (CS-B 12sh) and chromosome 26 (CS-B 26Lo) for different fibre traits. For example, the negative additive genetic effect for MIC with CS-B 01 and positive additive genetic effect for LP with CS-B 12sh, not detected in previous studies (Lacape et al. 2005; Zhang et al. 2005), could be useful in upland cotton improvement. Results also confirmed some of the challenges and difficulties associated with upland cotton improvement showing contrasting effects for some of the desirable traits within the same chromosome.

Our results suggest that some of the four CS-B lines may not have much to offer for the improvement of agronomic or fibre traits compared with improved upland cultivars. However, these CS-B lines were in a TM-1 background, a line with poor breeding value. Parental line TM-1 was chosen for developing these cytogenetic stocks because it is considered as the genetic standard line of upland cotton (Kohel et al. 1970; Kohel et al. 2001). Parental line 3-79 was used as the donor parent because it is a doubled haploid and is largely absent of genes introgressed from *G. hirsutum* (personal communication with Dr R.J. Kohel, USDA/ARS, TX, USA). 3-79 has poor agronomic qualities but superior fibre qualities.

Introgressing selected 3-79 genes from specific chromosomes or chromosome arms, via the CS-B lines, should reduce the amount of genetic drag associated with the poor agronomic qualities of *G. barbadense*. Genetic drag is a common problem associated with conventional interspecific crosses in upland cotton. Previous studies have documented that the genetic transmission in backcrosses of *G. hirsutum* with *G. barbadense* favours elimination of the donor genotype (*G. barbadense*) preserves genetic integrity of the recurrent parent and inhibits introgression of donor alleles (Stephen 1949, Jiang et al. 2000).

Our results from chromosomal assignment of QTL are in general agreement with other published map information (Rong et al. 2004, Lacape et al. 2005, Ulloa et al. 2005).

Previous studies have also reported the association of MIC and EL with chromosome 1 (Rong et al. 2004, Lacape et al. 2005). The long arm of chromosome 26 in line 3-79 which had a positive additive effect on SL and MIC but a negative additive effect on EL and T1 suggested that this specific region of the chromosome contributed significantly to fibre architecture. We also observed that the long arm of chromosome 26 in line 3-79 had high heterozygous dominance effect in many agronomic traits including BW, YLD and LY. Considering that 3-79 has smaller bolls and lower YLD and LY than TM-1 the results illustrate that the effect of some beneficial alleles are masked by some deleterious alleles in 3-79 and the CS-B lines could be useful to uncover the genetic effects of these beneficial novel alleles. A negative homozygous dominance effect for a CS-B parent might result in inbreeding depression in later generations. High heterozygous dominance effects between two parents might be associated with high heterosis in the F₁ or F₂ hybrid suggesting CS-B 26Lo could be useful in improving agronomic traits of hybrid cotton (Jenkins et al. 2006, 2007).

Ulloa et al. (2005) reported the chromosomal location of several major QTL for fibre and agronomic traits resided on the long arm of chromosome 26. They observed a very high density of molecular markers located on this chromosome arm, suggesting the association of a 'hot spot' of recombination.

Manipulating quantitative traits controlled by many genes located on 26 different chromosomes is difficult using conventional methods of interspecific breeding programmes for many reasons. Chromosome-specific introgression using CS-B lines can be used to overcome some of the problems encountered in conventional interspecific breeding programmes because these lines would reduce the genomic incompatibility between the two species (Jenkins et al. 2006, 2007).

The importance of these CS-B lines is that they help in the detection of individual chromosome or chromosome arm-specific genetic effects from the 3-79 genome and provide a tool for germplasm enhancement. Based on current estimates of the number of genes per eukaryotic plant genome, one would hypothesize that each substituted chromosome contains approximately 1500–2000 genes from *G. barbadense*. Even marker-assisted selection would not be able to detect and transfer all alien loci, especially in high recombination regions using conventional interspecific crosses. Development of chromosome substitution lines provides a unique opportunity to transfer all the genes associated with the introgressed alien chromosome or chromosome segment. The CS-B lines also provided a unique opportunity to discover novel alleles because of the unique genetic background of the CS-B lines. CS-B line evaluation does not always require segregating populations, so it differs from traditional QTL mapping and offers specific statistical advantages (Nadeau et al. 2000).

Currently, we are in the process of developing chromosome-specific recombinant inbred lines from crosses of individual CS-B lines with the recurrent TM-1 parent following the single seed descent (SSD) method. SSD allows high quality fibre traits from *G. barbadense* to be targeted for introgression into upland cotton, while reducing genetic drag associated with various agronomic traits of alien germplasm. Examples of transfer of genetic material from alien chromosomes using similar lines have been well documented in wheat (*Triticum aestivum*) and other plant species (Campbell et al. 2003, 2004,

Canady et al. 2005). CS-B lines will provide a valuable tool for genetic investigations and germplasm improvements in upland cotton.

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